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AB We present a simple yet powerful method for the isolation and analysis of exosomes released by antigen-presenting cells (APC). Exosomes are small vesicles (40-90 nm) released by APC, and may have an immuno-regulatory function in vivo. Such exosomes originate from MHC class II peptide loading compartments and, as such, express high levels of MHC Class II. We have utilised magnetic beads, coated with monoclonal antibodies specific for HLA DP, DQ, DR for the specific isolation of exosomes from cell-free supernatants. Beads coated with exosomes are subsequently stained with conjugated antibodies, and analysed by flow cytometry. Characterisation of exosomes by this method demonstrated that exosomes derived from B-lymphocytes express abundant MHC Class I and II molecules. Other immunologically important molecules detected included the co-stimulatory molecules B7.1 (CD80) and B7.2 (CD86). The adhesion molecule ICAM-1 (CD54) was also detected. These exosomes also expressed the B cell marker CD20, and the complement inhibitory protein CD59. The expression of CD63, a lysosomal marker, was variable, and there was no detectable expression of transferrin receptor (CD71). Monocyte derived dendritic cells (cultured for 7 days in GM-CSF/IL-4), demonstrated an immature phenotype, and secreted exosomes with a similar phenotype, with abundant MHC molecules. The expression of CD63 was consistently strong, and the MHC Class I-like molecule CD1a was also present, suggesting a possible function in the presentation of lipid antigens. Again CD59 was expressed suggesting a possible role for APC exosomes in complement regulation. There was no detectable CD71, CD40, CD14, CD20 or CD83. Modification of the extraction protocol allowed a comparative analysis of exosome secretion under various conditions. Treatment of cells with calcium ionophore, or phorbol ester resulted in apparent increases in exosome release, while the phosphatidyl inositol 3-kinase inhibitor, wortmannin, reduced exosome secretion. The immuno-magnetic isolation and analysis of exosomes is a versatile and rapid tool for the analysis of APC exosomes, and may prove a valuable tool for the study of exosome biology.